DERIVATIVES OF OXETANOCIN: OXETANOCINS H, X AND G, AND 2-AMINO-OXETANOCIN A

Sir:

Oxetanocin is a novel nucleoside isolated from a culture filtrate of *Bacillus megaterium* NK84-0218. It is the first natural product having an oxetanosyl-N-glycoside. Recently, the potential usefulness of OXT-A as an antiviral agent was disclosed by Hoshino et al. Therefore, we have studied chemical and biological transformation of OXT-A to get its purine nucleoside analogues. In this communication, preparation of oxetanocins H, X and G, and 2-amino oxetanocin A, and their preliminary biological activities are described. The detailed biological activities will be published in separate papers.

The scheme of chemical and biological transformation of OXT-A is shown in Fig. 1.

Oxetanocin H (OXT-H, the H comes from hypoxanthine) was readily derived from OXT-A by treatment with commercially available adenosine deaminase (Sigma, EC 3.5.4.4). MP 210°C; [α]D = -9.1° (c 1.0, H2O); field desorption mass spectra (FD-MS) m/z 252 (M+), calc for C16H12N4O4: C 47.62, H 4.80, N 22.21, found: C 47.41, H 4.96, N 22.05; UV λmax nm (log ε) 248.5 (4.10); λmax HCl 249 (4.08); λmax NaOH 254 (4.12).

For the microbial transformation, we tested 48 microorganisms including bacteria, actinomycetes, fungi and yeasts. The freshly harvested cells or mycelia were suspended in 1/20 M phosphate buffer (pH 7.0) containing OXT-A at 50 μg/ml and incubated for 18 hours at 37°C (for bacteria) or for 48 hours at 28°C (for other microorganisms). The transformation was analyzed by HPLC [column, Nucleosil 5C-18 (4.6×250 mm); mobile phase, 0.1 M citrate buffer (pH 4.0) - CH3CN - MeOH (50:2:1); flow rate, 0.7 ml/minute; temp, 21°C; detection, UV at 259 nm]. The retention times for OXT-A and OXT-H were 17’30” and 9’20’, respectively. Among the 48 microorganisms, 9 microorganisms including *Escherichia coli* 120551 converted OXT-A to OXT-H almost quantitatively.

In these studies we found that some microorganisms belonging to actinomycetes including *Nocardia intermedia* converted OXT-A to another nucleoside (retention time, 11’5”) via OXT-H.

Fig. 1. Chemical and biological transformation of oxetanocin A.

(a) Adenosine deaminase or *Escherichia coli* 120551. (b) *Nocardia intermedia* (from OXT-A against OXT-H). (c) Three step chemical reaction (see Fig. 2). (d) Adenosine deaminase.

- Hereafter, oxetanocin is named as oxetanocin A (abbreviation: OXT-A). The A comes from adenine chromophore.
This new nucleoside was found to be oxetanocin X (OXT-X, the X comes from xanthine). MP 205°C (dec); [α]_D^22 -22.0° (c 0.64, H₂O); FD-MS m/z 269 (M+H)+, calcd for C₁₀H₁₂N₄O₅·H₂O: C 41.96, H 4.93, N 19.57, found: C 41.84, H 4.95, N 19.43; UV λmax nm (log ε) 250.5 (4.01), 276.5 (3.95). The conversion from OXT-A to OXT-X by N. interforma was almost quantitative.

We were successful in the chemical transformation of OXT-A to 2-amino-OXT-A, which is readily transformed to OXT-G (G from guanine), via the 2,6-dichloropurine derivative, but the overall yield was only 7.2%. Therefore, we tried microbial transformation from OXT-X to OXT-G, but this attempt was not successful. The effective transformation was achieved by three step chemical reaction followed by enzymic deamination. The scheme of the chemical transformation is shown in Fig. 2. Treatment of OXT-X with acetic anhydride and triethylamine in the presence of a catalytic amount of 4-dimethylaminopyridine gave the di-O-acetate in 90% yield. It was treated with 2,4,6-triisopropylbenzenesulfonyl chloride and triethylamine in the presence of 4-dimethylaminopyridine to yield the di-O-sulfonyl derivative in 90% yield. The ammonolysis of the product at 110°C for 3 days in a pressure vessel gave 2-amino-OXT-A in 56% yield. MP 112°C (dec); [α]_D^22 -19.8° (c 0.75, 0.1 N NaOH); FD-MS m/z 268 (M+H)+, calcd for C₁₀H₁₃N₅O₄·H₂O: C 42.10, H 5.30, N 24.55, found: C 42.04, H 5.18, N 24.48; UV λmax nm (log ε) 253.5 (4.09).

All of the new derivatives presented in this paper did not show antibacterial activities except for 2-amino-OXT-A: Staphylococcus aureus 209 P (MIC: 3.13 μg/ml in 0.5% peptone agar). Bacillus cereus IAM 1072 (3.13 μg/ml).

Antiviral activities against herpes simplex virus type-II (HSV-II) are shown in Table 1. Antiviral activities against human immunodeficiency virus etc. will be presented in separate papers.

Acknowledgments

The authors are indebted to Mr. S. Inada for the FD-MS spectrometry, and to Mr. K. Matsuo and Mrs. K. Aoyama for the antiviral activity.
Nobuyoshi Shimada
Shigeru Hasegawa
Seiichi Saito
Takaaki Nishikiori
Akio Fujii
Tomohisa Takita

Research Laboratories,
Pharmaceutical Group,
Nippon Kayaku Co., Ltd.,
3-31-12 Shimo, Kita-ku,
Tokyo 115, Japan

(Received July 25, 1987)

References